

CHEMICAL EVALUATION OF THE GENUS *ACACIA* OF PAKISTAN
Part I. Fatty Acid Composition of the Total Lipids and Lipid Classes of *Acacia albida* Del.
and *Acacia modesta* Wall.

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Acacia albida Del. seed oil (3.64%) and *Acacia modesta* (6.5%) were examined for their fatty acids composition. The oils were fractionated by TLC into lipid classes; which were neutral lipids (39.0, 55.3%) and polar lipids (60.9%, 44.7%) respectively. Further fractionation of the neutral lipids resulted hydrocarbons, wax-esters, triglycerides, free fatty acids, 1:3-diglycerides, 1:2-diglycerides and monoglycerides. Fatty acids composition of the whole oil and its fractions were determined by GC. This is the first report of the detection of margaric acid in *Acacia albida* seed oil.

Key words: Leguminosae, Genus *Acacia*, Lipid composition.

INTRODUCTION

Acacia is a large genus of the Leguminosae family consisting of about 800 species. Most of the species are native to Pakistan, India, Australia, Africa and America [1]. The *Acacia* plants can grow under the most severe conditions of temperature, soil and climate. Thirty four species have been reported to grow in Pakistan. Among these some are cultivated, others grow wild [2,3].

Acacia albida Del. is a native of tropical and subtropical Africa. In Pakistan, it has been introduced in Kurram Valley (NWFP) area and is scattered in Punjab also. In African countries, a decoction of its bark is taken twice a day as a cure of cough and diarrhoea [4].

The chemical analysis of *Acacia albida* seeds showed the presence of α amino β oxalylamino-propionic acid which is a toxic amino acid and causes neurolathyrism in man and in cattle [5,6]. Grindley [7] reported the fatty acid composition of the seed oil alongwith some other Sudanese *Acacia* seed oils. These *Acacia* seed oils contained oleic acid, linoleic acid, saturated acids (mainly palmitic) and higher saturated acids. Gunstone *et al* [8] also examined the *Acacia albida* seed oil of the zambian variety. They found palmitic, oleic and linoleic acids as the principal component fatty acids of the seed oil.

Acacia modesta Wall. is locally known as 'Phulai'. The plant is widely distributed in Pakistan, India and Afghanistan [1]. The seeds have been reported to have hypoglycemic activity [9].

The chemical analysis of *Acacia modesta* has been first ever reported in the literature by Joshi *et al* [10]. They reported the presence of α -amyrin, betulin, octacosanol and β -sitosterol in C_6H_6 extract of the stem bark of

the species. The petroleum ether and alcoholic extracts of the heartwood of the plant gave octacosane, hentriacontane, octacosanol and hentriacontanol.

No work has been reported on the seed oil of *Acacia albida* of the variety of Pakistan, and the seed oil of *Acacia modesta*. In the present investigations, seed oils have been examined for their fatty acid composition as a whole and their different lipid fractions for their exploitation as for shortenings substitute or otherwise for soap industry. The studies are in continuation of our previous work for the exploration of new sources of edible or commercially important lipids.

MATERIALS AND METHODS

The seeds of *Acacia* species were collected from around the Lahore District, Pakistan and were finely ground. The ground seeds were extracted with chloroform: methanol (2:1) solvent mixture according to the procedures of Folch *et al* [11]. Non lipid contents were removed by the use of salt solution [12]. The extract was then dried over anhydrous sodium sulphate. The solvent was removed in a rotary evaporator under vacuum to yield the oil.

The portion of the oil was fractionated into lipid classes by the use of thin layer chromatography [18]. The lipid fractions such as wax-esters, triglycerides, free fatty acids, monoglycerides, 1:2-diglycerides, 1-3-diglycerides and polar lipids were eluted and separated. The corresponding lipid fractions were extracted from the silica gel, the solvent was removed and their weight percentages were determined (Table 1).

Methyl esters of the whole oil and each lipid fraction except the hydrocarbons were prepared by the use of borontri-fluoride solution in methanol [17]. The esters were analyzed for their fatty acid composition by GC on a Pye Unicam 204 Series unit equipped with a flame ion-

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ization detector and a 4 mm x 1.5 m glass column packed with 10% diethylene glycol succinate on diatomite CAW. The column was maintained at 200^o, and nitrogen was used as the carrier gas at a flow rate of 40 ml/min. The peaks thus obtained were identified by comparison of their retention times with those of the standard methyl esters under the same conditions. Percent peak areas are quoted as composition percent weight in Table 3 and 4.

RESULTS AND DISCUSSION

The lipid contents of *Acacia albida* and *Acacia modesta* seeds, their classes and fatty acids composition have been shown in Table 1, 2, 3 and 4. The total lipids in *Acacia albida* and *Acacia modesta* were found to be 3.64% and 6.5% respectively. The purified lipids contained higher amount of neutral lipids (55.3%) than the polar lipids (44.7%) in *Acacia modesta* whereas the purified lipids of *Acacia albida* contained higher amount of polar lipids (60.9%) than the neutral lipids (39.0%).

The neutral lipids were further fractionated by thin layer chromatography and the classes identified and collected were hydrocarbons, wax esters, triglycerides, free

fatty acids, 1,3-diglycerides, 1,2-diglycerides and monoglycerides (Table-1). The analysis of polar lipids fraction is in progress but the fatty acid composition of the total lipid fraction was carried out in order to make the comparison of the fatty acid composition of both the species and the results are recorded in Table 2 and 3. The seed oil of *Acacia modesta* is rich in saturated fatty acids and the predominant unsaturated fatty acid is oleic whereas the major fatty acids present in the seed oil of *Acacia albida* were palmitic, oleic and linoleic. Although the fatty acid composition of many species of *Acacia* have been reported in the literature [13]; it is for the first time that the fatty acid composition of the *Acacia albida* seed oil has been examined. Fatty acid composition of the *Acacia modesta* seed lipid is quite comparable to the findings of Chaudhry *et al* [13] who studied seven *Acacia* seed oils i.e. *Acacia auriculiformis*, *A. catechu*, *A. coriacea*, *A. farnesiana*, *A. mellifera*, *A. miuhassai*, *A. nilotica* and *A. penata* and found oleic and linoleic as the predominant fatty acids in all these oils. They also reported that all these oils were rich in unsaturated fatty acids. Mannan *et al* [14] analysed the seed oil of *Acacia mellifera* and found palmitic, oleic and linoleic to be the major fatty acids. They also reported the presence of lauric acid as the minor component.

Table 1 Percentage of lipid fractions of *Acacia* species.

Lipid fraction	Percent	
	<i>Acacia albida</i>	<i>Acacia modesta</i>
Neutral	39.0	35.0
Polar	60.9	44.7
Hydrocarbons	2.1	2.6
Wax esters	2.0	6.0
Triglycerides	20.7	28.1
Free fatty acids	4.3	3.0
1:3-Diglycerides	3.5	6.8
1:2-Diglycerides	2.6	2.9
Monoglycerides	3.8	5.9

The composition of seed oil fractions of *Acacia albida* showed that the major fatty acid present were palmitic, oleic and linoleic acids. Grindley *et al* [7] also reported palmitic, oleic and linoleic acids as the predominant fatty acids, whereas Gunstone *et al* [8] reported the presence of palmitic, palmitoleic, stearic, oleic linoleic, linolenic, arachidic and behenic acids in the seed oil of the Zambian variety. However, our results showed the presence of other acids such as lauric, myristic and an odd numbered fatty acid C 17:0 (margaric acid), as well.

It is the first ever report of the presence of margaric acid in the fatty acid composition of the *Acacia albida* seed oil of the Pakistan. Choudhry [13] as well as Shhina *et al* [15] studied the fatty acid composition of some of the *Acacia* species. They did not report the presence of mar-

Table 2 Percent fatty acids composition of total lipids of *Acacia* species.

Species	Source	Lipids %	Component ester, we. %													
			10:0	12:0	14:0	16:0	16:1	17:0	FAI	18:0	18:1	18:2	20:0	18:3	FAI	22:0
<i>Acacia albida</i>	Zambian	5.0	—	—	—	19.0	1.0	—	—	3	23	51	1.0	1.0	—	1.0
<i>Acaica albida</i>	Pakistani	3.6	—	0.96	1.23	21.92	2.74	1.78	1.64	8.9	28.77	18.63	24.6	Traces	6.16	4.79
<i>Acacia mqdesta</i>	Pakistani	6.5	3.8	3.58	4.94	21.14	—	—	—	19.42	26.30	12.15	—	8.66	—	—

Table 3. Percent fatty acids composition of lipid classes of *Acacia albida* Del.

	C _{12:0}	C _{14:0}	C _{16:0}	C _{16:1}	C _{17:0}	FAI	C _{18:0}	C _{18:1}	C _{18:2}	C _{20:0}	C _{18:3}	FAII	C _{22:0}	Percent saturated fatty acids	Percent unsaturated fatty acids
Wax esters	4.72	13.48	16.17	10.78	4.98	9.43	15.50	9.16	8.08	3.90	Traces	2.42	1.35	60.10	28.02
Triglycerides	0.74	1.27	20.38	2.55	1.40	1.66	8.92	30.58	24.48	3.18	Traces	Traces	3.82	39.71	58.61
Free fatty acids	Traces	0.64	21.94	1.91	1.53	1.27	12.75	30.61	20.40	3.19	1.91	Traces	3.83	43.88	54.83
1:3-Diglycerides	0.68	1.53	27.21	4.76	4.25	3.93	10.37	24.49	7.48	4.76	3.06	2.38	5.10	53.90	39.79
1:2-Diglycerides	0.54	0.90	28.83	2.16	4.50	1.47	10.99	32.43	5.40	3.06	2.70	3.78	3.24	52.06	42.69
Monoglycerides	1.10	1.38	22.16	4.71	4.15	4.02	11.08	33.24	4.43	3.36	Traces	6.65	3.60	46.93	42.38
Polar lipids.	1.54	2.05	34.94	3.70	3.70	4.11	10.28	19.53	2.05	2.46	Traces	11.10	4.52	59.49	25.28

Table 4. Percent fatty acids composition of lipid classes of *Acacia modesta*

Fractions	C _{10:0}	C _{12:0}	C _{14:0}	C _{16:0}	C _{18:0}	C _{18:1}	C _{18:2}	C _{18:3}	% Saturated acids	% Unsaturated acids
Wax ester	1.22	5.17	8.34	17.98	17.88	23.85	15.32	10.23	50.59	49.40
Triglycerides	Traces	3.38	4.28	24.21	8.14	26.03	24.82	9.13	40.01	59.98
Free fatty acids	6.12	4.08	5.10	24.48	10.23	18.36	22.44	9.18	50.01	49.98
1,3-Diglycerides	1.59	3.48	4.99	23.88	5.96	26.26	23.88	9.95	39.90	60.09
1,2-Diglycerides	2.17	4.46	4.90	19.28	8.87	23.55	26.48	10.28	39.68	60.31
Monoglycerides	3.08	4.24	5.19	20.61	9.68	25.48	22.79	8.92	42.80	57.19
Polar lipids	Traces	4.86	4.05	24.81	12.40	28.19	15.35	10.33	46.12	53.87

garic and behenic acids in the seed oils of the species investigated by them. But, Choudhry [13] reported the presence of an odd numbered saturated fatty acids C 15:0 in the seed oil of *A. pennata* of Indian origin. Two more fatty acids were detected and could not be identified by us. The investigations are under progress. The results of other workers [15,16] on different *Acacia* species are comparable to our results. Our studies showed that the *Acacia* seed

oils have C_{16:0} C_{18:1} and C_{18:2} as the principal fatty acids.

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